

**HEPATOZOON TACHYGLOSSI SP. NOV. (HAEMOGREGARINIDAE),  
A PROTOZOAN PARASITE FROM THE BLOOD OF A SHORT-BEAKED ECHIDNA,  
TACHYGLOSSUS ACULEATUS**

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**Summary**

CLARK, T., HOLZ, P. & SPRATT, D. M. (2005) *Hepatozoon tachyglossi* sp. nov. (Haemogregarinidae), a protozoan parasite from the blood of a short-beaked echidna, *Tachyglossus aculeatus*. *Trans. R. Soc. S. Aust.* 129(1), 49–52, 31 May, 2005.

*Hepatozoon tachyglossi* sp. nov. is described from monocytes in the peripheral blood of a debilitated short-beaked echidna from the Healesville region of Victoria. Of the *Hepatozoon* Miller, 1908 species known to occur in Australian native mammals, all of those in marsupials occur in erythrocytes and only *H. muris* from introduced and native rodents occurs in monocytes. *H. tachyglossi* is distinguished from *H. muris* by its larger size and the lack of a capsule.

KEY WORDS: *Hepatozoon tachyglossi*, new species, echidna, *Tachyglossus aculeatus*.

**Introduction**

The blood of the short-beaked echidna, *Tachyglossus aculeatus* (Shaw & Nodder, 1792), has yielded relatively few haemoparasites. Those identified have been restricted to the Piroplasmidae and include *Babesia tachyglossi* (Backhouse & Bolliger, 1959) and *Theileria tachyglossi* (Priestly, 1915) (Backhouse & Bolliger 1957, 1959; Mackerras 1959). We report a novel species of *Hepatozoon* from monocytes in the peripheral blood of a short-beaked echidna from the Healesville region of Victoria and compare the species with other members of the genus previously described from Australian native mammals.

**Clinical history and methods**

A male, juvenile, short-beaked echidna was presented to the veterinary service of Healesville Sanctuary. The animal had several injuries including a damaged tongue and a fractured humerus. During captive management and treatment, the animal developed severe dyspnoea and a mucopurulent nasal discharge. It was euthanased after a poor response to treatment with antimicrobial drugs and supportive therapy.

A sample of blood was collected ante-mortem from the bill sinus. The morphology of haematological cells in a blood film, stained with Wright's and Giemsa stains, was assessed by light microscopy. Tissue

samples of bone marrow, liver and spleen, were collected post-mortem and fixed in 10 percent buffered formalin. The tissues were processed using standard histological methods; sections were stained with haematoxylin and eosin stains and examined using light microscopy. The organisms and their host cells were digitally photographed. A stage micrometer was photographed at the same magnification and used to insert scale bars to all micrographs. The micrometer was also used for all measurements which are presented in microns as the mean  $\pm$  standard deviation followed by the range in parentheses.

**Results**

Intracellular organisms were observed within leukocytes in the peripheral blood (Fig. 1). These were observed in 24/50 monocytes but were not observed in any granulocytes.

*Hepatozoon tachyglossi* sp. nov.  
(FIG. 1)

*Host*

*Tachyglossus aculeatus*.

*Location*

Healesville, Victoria.

*Type*

Slide in South Australian Museum No. 28751.

Typically the organisms were oval to elongate in shape but were quite pleomorphic with some pyriform and round forms observed. The organisms were  $9.9 \pm 1.4$  (7.8–12.4) in length and  $4.7 \pm 0.7$  (3.8–5.9) in width ( $n = 18$ ). Most organisms had an eccentric, subterminal nucleus and some exhibited a

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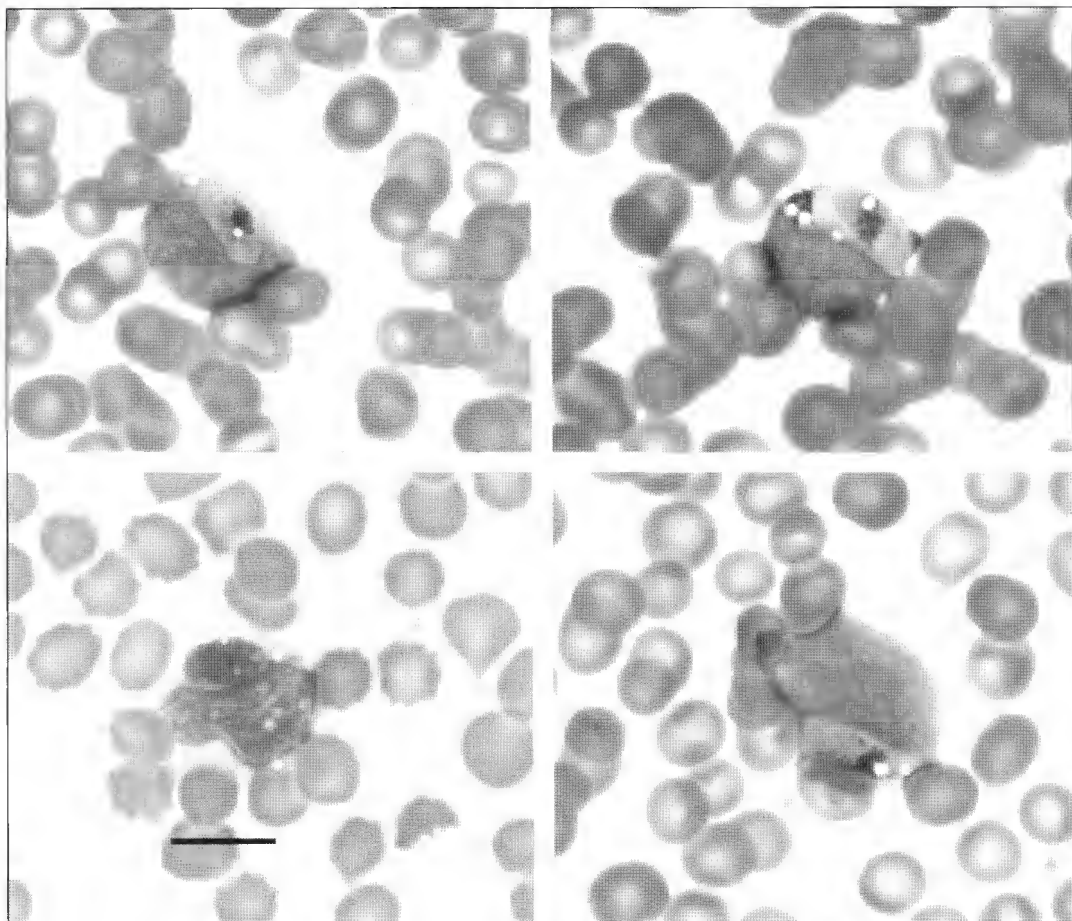


Fig. 1. Examples of *Hepatozoon tachyglossi* sp. nov. within monocytes from the peripheral blood of a short-beaked echidna. Bar = 10  $\mu$ m.

small amount of punctate, dark brown – black pigment in the cytoplasm. No distinct capsule was observed and there was only a subtle difference in colour between the cytoplasm of the organism and the cytoplasm of the cell. Only one organism was evident per cell; in some cases this caused displacement of the host cell nucleus. No consistent changes in cell morphology were evident. No extracellular organisms were noted. Examination of histological sections of bone marrow, liver and spleen did not reveal schizonts.

### Discussion

The organism described is morphologically similar to a species of *Hepatozoon* Miller, 1908, the only coccidian genus that inhabits the blood of mammals. Desser (1990) highlighted the taxonomic confusion

arising from the problem of differentiating species of *Hepatozoon* from that of *Haemogregarina* Danilewsky, 1885 on the basis of gamonts in the blood of the vertebrate host. Differentiation was based primarily on the size of the oocysts and the presence or absence of sporocysts in the invertebrate definitive host. Desser (1990) tested and confirmed the hypothesis of Landau *et al.* (1972) that tiny cystic stages (cystozoites) in the liver and lungs represent a common feature in all species of *Hepatozoon* and, as a consequence, differentiation of the genera *Hepatozoon* and *Haemogregarina* based solely on the stages in the vertebrate host is possible. These findings further support the evidence of Landau *et al.* (1972) that transmission of species of *Hepatozoon* may be by predation, as well as by ingestion of the infected arthropod vectors.

The genus has not been reported previously in

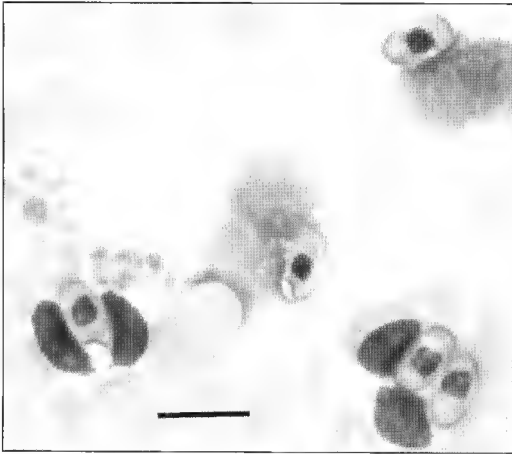


Fig. 2. Examples of *Hepatozoon muris* from the blood of *Rattus lutreolus*. Bar = 10  $\mu$ m.

monotremes. However, *H. peramelis* (Welsh & Dalyell, 1909), *H. dasyuroides* Mackerras, 1959, *H. dasyuri* (Welsh, Dalyell & Burfitt, 1909), *H. petauri* (Welsh & Barling, 1909) and *H. pseudocheiri* Mackerras, 1959, have been described from Australian marsupials (see Mackerras, 1959). In addition, O'Donoghue and Adlard (2000) reported several unidentified species of *Hepatozoon* from Australian marsupials. All reports have recorded the parasite within erythrocytes or 'free' in the blood (Mackerras 1959; Speare *et al.* 1984; Bettiol *et al.* 1996). Also, *H. muris* (Balfour, 1906) has been recognised in both the introduced *Rattus norvegicus* (Berkenhout, 1769) and *R. rattus* (Linnaeus, 1758), and the native rodents, *R. fuscipes* (Waterhouse, 1839) and *R. sordidus* (Gould, 1858) (Mackerras 1959; O'Donoghue & Adlard 2000). *Hepatozoon muris*, in contrast to the species reported in marsupials, infects leukocytes (Soulsby 1982).

The organisms in the current study were oval to elongate in shape, as are many species of *Hepatozoon*, and were similar in size to *H. peramelis* (9.0 – 10.0  $\mu$ m by 3.0 – 3.5  $\mu$ m; Welsh & Dalyell

1909) and slightly larger than *H. petauri* (7.5 – 8.0  $\mu$ m by 3.5 – 4.0  $\mu$ m; Mackerras 1959), *H. pseudocheiri* (8.0 – 13.0  $\mu$ m by 1.5–3.0  $\mu$ m; Mackerras 1959), and *Hepatozoon sp.* (8.7  $\pm$  0.2  $\mu$ m by 2.1  $\pm$  0.4  $\mu$ m, Bettiol *et al.* 1996). In contrast, *H. dasyuroides* (12 – 13  $\mu$ m by 1 – 2  $\mu$ m; Mackerras 1959) and *H. dasyuri* (12  $\mu$ m by 4  $\mu$ m; Welsh Dalyell & Burfitt 1909, 1910) are longer, narrower parasites.

*Hepatozoon tachyglossi* sp. nov., like *H. muris*, occurs in the monocytes of its host. However, *H. muris* is a smaller (7.0 – 8.0  $\mu$ m by 3.0 – 3.5  $\mu$ m; Mackerras, 1959) more ovoid, and typically less morphologically variable parasite, than *H. tachyglossi* (Fig. 2). Additionally, *H. muris* has an eosinophilic capsule that was not evident in *H. tachyglossi*.

Typically, the definitive hosts and vectors of species of *Hepatozoon* are blood feeding arthropods. The mite *Laelaps echidninus* Berlese, 1887 fills this role for *H. muris*. In the current case, ticks identified as *Aponomma concolor* Neumann, 1899, were evident on the echidna but were not examined for sporocysts and could not be proven to be the definitive host.

Although the life history of this species of *Hepatozoon* remains unknown, we consider that specific status is warranted on the basis of morphological features and the occurrence of the parasite in monocytes of the monotreme, *Tachyglossus aculeatus*.

The effect on the host was not determined due to the other pathological processes and the animal's debilitated state may have allowed the organism to proliferate. Further work to identify the prevalence of this organism in short-beaked echidnas, assess any pathogenic effects on the animal and phylogenetic studies to determine its relationship to other species of *Hepatozoon* in Australia need to be undertaken.

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